



IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant : Richard T. Dean et al.

Art Unit : 1654

Serial No.: 08/236,402

Examiner : Jeffrey Russel

Filed : May 2, 1994

Title : TECHNETIUM-99M LABELED IMAGING AGENTS

**DECLARATION UNDER 37 C.F.R. § 1.132**

I, JOHN LISTER-JAMES, declare as follows:

1. I reside in Bedford, New Hampshire.

2. I received my B.Sc., with honors, in chemistry from Imperial College of the University of London in 1974, and I was awarded my Ph.D. in organic chemistry from the University of London in 1981. From 1981 to 1986 I was a Research Fellow in Radiology (Nuclear Medicine), Children's Hospital, Boston, Massachusetts. During the same time I was a Visiting Scientist at the Massachusetts Institute of Technology, Cambridge, Massachusetts. From 1983 to 1986 I was also an Associate in Radiology (Nuclear Medicine), Harvard Medical School, Boston, Massachusetts. During this period, my research included the design and synthesis of technetium complexes, the design, synthesis and evaluation of <sup>99m</sup>Tc-labelled radiopharmaceuticals, the investigation of structure/activity relationships probing biochemical processes and the development of new methods in organic synthesis. From 1987 to 1990 I was employed at Centocor, Inc., Malvern, Pennsylvania, in various senior positions. At Centocor, I supervised the product development support for, wrote sections of, and coordinated the completion of the technical sections of US and European Product License Applications for a radioimmunodiagnostic product. I also supervised technical projects supporting Product License Application submissions, and I supervised a group of senior and associate

scientists in the development of novel protein modification and radiolabelling methods leading to proprietary and potentially commercially valuable technology. From 1990 to 1999, I was employed by Diatide, Inc. (formerly Diatech, Inc.) in various management positions, including Director of R&D, Director of Regulatory Affairs, Director of Product Development, Senior Director of R&D, Vice President of Science & Technology. During this time I was responsible for various aspects of radiopharmaceutical R&D, in particular of radiopharmaceuticals based upon technetium labeled peptides. At different times during this period I was responsible for drug discovery, intellectual property, product development, preclinical pharmacology and toxicology and regulatory affairs. In 1999, Berlex Laboratories, Inc. acquired Diatide, Inc., and since 1999 I have been employed by Berlex. I am currently Vice President, Research & Development, Berlex Diagnostic Imaging and Radiopharmaceuticals. I am currently responsible for a research and development group involved in radiopharmaceutical drug discovery, chemistry, manufacturing and controls development, and clinical development of radiopharmaceuticals.

3. I am an inventor in the 58 patents set forth in the attached Exhibit 1 and an author of numerous scientific articles, including those listed in Exhibit 2 attached hereto.

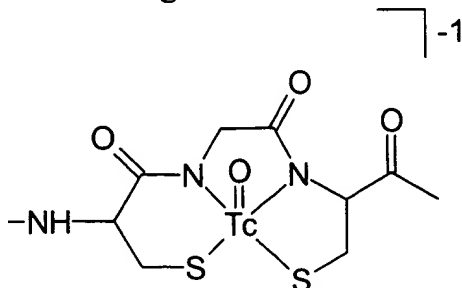
4. I am also an inventor in 2 recently issued United States Patents shown in the attached Exhibit 2.

5. I have been asked to review and comment on certain references cited by the Examiner in rejecting various claims in this patent application. In particular, I am submitting this declaration in support of the concept that technetium will bind to an -S-N-N-S- chelator in preference to an -N-N-N-S- chelator.

6. Technetium is most readily available in the +7 oxidation state as pertechnetate. In order to cause it to bind to other molecules, the technetium must be reduced to a lower oxidation state. The first and most accessible reduced form of technetium is the +5 oxidation state, formed by a two-electron reduction of pertechnetate. The most common two-electron reducing agent used in technetium-99m-based radiopharmaceuticals is stannous ion in the form of a stannous salt such as stannous chloride, although other reductants can be used. In order to radiolabel a specific-binding molecule with the reduced technetium, the technetium must be bound as a coordination complex with a ligand that is part of, or covalently linked to, the specific-binding molecule.

7. In its +5 oxidation state, technetium normally forms square pyramidal complexes with one oxo ligand and 4 other donor ligands. In this form, technetium is thiophilic, preferring thiolate, *i.e.*, sulfur (+2), donor ligands. This was exemplified in an article by DePamphilis et al. – Bruno V. DePamphilis, Alun G. Jones and Alan Davison, "Ligand-Exchange Reactivity Patterns of Oxotechnetium(V) Complexes", Inorg. Chem. 22:2292-2297 (1983) – copy attached as Exhibit 3. The authors reported that, when reacted with 2-mercaptoacetic acid (a/k/a thioglycolic acid) having the molecular structure HSCH<sub>2</sub>COOH, technetium in the +5 oxidation state preferentially formed a bis-(2-mercapto-thiolacetic acid) complex with the 2-mercapto-thiolacetic acid (molecular structure HSCH<sub>2</sub>COSH) present as an impurity in the 2-mercapto-acetic acid.

8. In addition, it has been shown that stable oxo complexes of technetium (+5) can be formed from bisamide bithiol ligands.



See, Alan Davison, Alun G. Jones, Chris Orvig and Miriam Sohn, "New Class of Oxotechnetium (5+) Chelate Complexes Containing a  $\text{TcON}_2\text{S}_2$  Core", Inorg. Chem. 6:1629-1632 (1981) – copy attached as Exhibit 4. In Dean et al. U.S. Patent No. 5,561,220, and in Dean et al. U.S. Patent No. 6,017,510, the inventors have claimed compositions for radiolabelling specific binding peptides using such ligands presenting -Cys-amino acid-Cys- sequences (with the cysteine thiol groups initially in technetium-cleavable protected form).

9. Therefore, it is expected that, when presented with a molecule containing more than one thiol donor ligand, technetium will bind preferentially to both thiols when there are two available, or to as many as four thiols if present and configurationally possible.

10. Although it is now known that technetium (+5) can form stable oxo complexes with single thiol containing ligands, such as mercaptoacetyl-Gly-Gly-Gly, the known thiophilicity of technetium would cause one to expect that, when more than one thiol group is available for binding, technetium will form a complex across two or more thiol groups.

11. Therefore, in a peptide sequence such as peptide -Gly-Gly-Cys-Gly-Cys-Gly-Gly-peptide, it would be expected that technetium will form an oxo complex with the -Cys-Gly-Cys- sequence, in preference to forming a complex with either the -Gly-Gly-Cys- or -Cys-Gly-Gly- sequences. This is even more likely considering that, for bisamide bithiol complexes of technetium (+5), ligands that form 5 or 6 membered rings with the technetium are preferred (Exhibit 4). The sequence -Cys-Gly-Cys- is a ligand of this type.

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I declare further that all statements made of my own knowledge are true and all statements made on information and belief are believed to be true. I make this declaration understanding that willful false statements and the like are punishable by fine or imprisonment, or both, under 18 U.S.C. § 1001 and that such willful statements may jeopardize the validity of this application or any patent issuing thereon.

Signed at Londonderry, New Hampshire, this      day of June 2003.

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John Lister-James

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